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Address for correspondence: Heping Xiao, Department of Tuberculosis, Shanghai Pulmonary Hospital, Tongji University School of Medicine, No. 507 Zhengmin Rd, Shanghai, People's Republic of China 200433; email: xiaoheping_sars@163.com



## Clade 2.3.2 Avian Influenza Virus (H5N1), Qinghai Lake Region, China, 2009–2010

**To the Editor:** In 2005, a large population of wild migratory birds was infected with highly pathogenic avian influenza (HPAI) virus (H5N1) in the Qinghai Lake region of western People's Republic of China, resulting in the death of ≈10,000 birds (*1*,*2*). On the basis of phylogenetic analysis of the hemagglutinin (HA) gene, the virus was classified as clade 2.2 according to the World Health Organization guidelines. Subsequently, viruses from this clade were found in Mongolia, Russia, Europe, and Africa along the migratory flyways of birds (*3*,*4*). This unique distribution of the same clade of HPAI virus (H5N1) through different migratory routes indicates that migratory birds might play a global role in virus dissemination (*3*,*4*).

In 2006, viruses from the same clade were isolated in the Qinghai Lake region (*3*). Analysis of viral outbreaks along migratory flyways demonstrated a similar outbreak pattern for the past 4 years (2006–2009) (*5*). During that period, clade 2.2 avian influenza virus (H5N1) was isolated in China, Mongolia, Russia, Germany, Egypt, and Nigeria; all viruses were closely related to the Qinghai Lake virus. Despite the broad distribution of clade 2.2 viruses in migratory flyways, few isolates of clade 2.2 viruses in local domestic poultry were reported, especially in China (*6*). Outbreaks of these viruses were reported in poultry in Africa (*7*). The reason these viruses rarely cause outbreaks in poultry is unknown.

During May–June 2009 and 2010, several dead migratory birds were found in the Qinghai Lake region. Nine HPAI viruses (H5N1) were isolated in 2009 and 2 were isolated in 2010 from great cormorants (*Phalacrocorax carbo*), brown-headed gulls (*Chroicocephalus brunnicephalus*), great black-headed gulls (*Ichthyaetus ichthyaetus*), great-crested grebes (*Podiceps cristatus*), and bar-headed geese (*Anser indicus*) and serotyped as described (*3*). HA genes from all 11 isolates were subsequently amplified by using reverse transcription–PCR and sequenced.

Phylogenetic analysis of HA sequences and an additional HA gene sequence from the 2009 Qinghai Lake subtype H5N1 virus isolate from a great crested grebe (from the National Avian Influenza Virus Reference Laboratory, Harbin, China) (GenBank accession no. CY063318) showed that HA genes from all 12 viruses clustered as clade 2.3.2 (Figure); none clustered with clade 2.2 viruses. Additionally, the HA cleavage site in the new isolates is PQRERRRKRG, which is identical to that of clade 2.3.2 viruses. In clade 2.2, the cleavage site is PQRERRRKKRG.

A bootstrap (1,000×) maximum likelihood tree (*8*) also demonstrated that Qinghai 2009 and 2010 virus isolates are closely related to those isolated in Mongolia and Uvs Nuur Lake in 2009, as reported by Sharshov et al. (*5*). Qinghai Lake and Uvs Nuur Lake, which are found along the migratory flyway in central Asia, are major lakes for bird migration and breeding. Many birds fly from Qinghai Lake to Uvs Nuur Lake in the spring.

If one considers isolation date and bird species infected, viruses isolated in Mongolia and Russia and our isolates were likely transmitted between the 2 lake regions by bird migration. Moreover, HA sequences are closely related to viruses isolated from wild birds in Hong Kong and Japan during 2007–2008, which are the most recent isolates of clade 2.3.2 viruses before isolation of 2009 Qinghai Lake viruses. These results indicate that viruses in the Qinghai Lake region may be transmitted by wild birds along the migratory flyway in eastern

Asia. However, there is no evidence that avian influenza virus (H5N1) is transmitted from eastern Asian (inner China or across the Himalayas) to the Qinghai Lake region.

The 2009 and 2010 Qinghai Lake viruses are related to various viruses isolated from plateau pikas near Qinghai Lake (*9*). In 2007, clade 2.2 and clade 2.3.2 viruses were isolated from plateau pikas, but no clade 2.3.2 viruses were found in aquatic birds. Wild birds, pikas, and other animals near Qinghai Lake share the same environment, and viruses may be transmitted across species. However, surveillance data are limited for wild animals near Qinghai Lake. Therefore, further investigations need to be conducted to clarify relationships among birds, animals, and influenza viruses near Qinghai Lake.

Our results and those of Sharshov et al. (*5*) show that in 2009 HPAI virus (H5N1) began infecting birds along the migratory route near Qinghai Lake and changed from clade 2.2 viruses to clade 2.3 viruses. New outbreaks of HPAI viruses (H5N1) along this migratory flyway should be investigated.

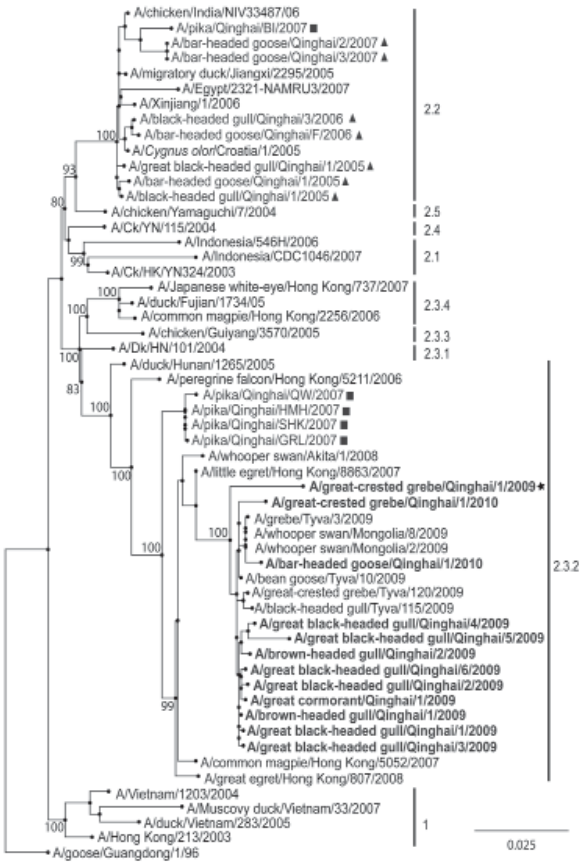


Figure. Bootstrapped (1,000×) maximum likelihood phylogenetic tree of hemagglutinin genes of avian influenza viruses (H5N1), People's Republic of China, 2009–2010. Viruses isolated from the plateau pika near Qinghai Lake are indicated by squares; viruses isolated from wild birds in Qinghai Lake Region during 2005–2007 are indicated by triangles; 2009 Qinghai virus submitted to GenBank by the National Avian Influenza Virus Reference Laboratory (Harbin, China) is indicated by the star and in **boldface**; and viruses isolated in 2009 and 2010 in the Qinghai Lake Region are indicated in **boldface**, Clade numbers are indicated on the right. NAMRU3, Naval Medical Research Unit 3; Ck, chicken; Dk, duck. Scale bar indicates nucleotide substitutions per site.

This study was supported in part by the China Ministry of Science and Technology Project 973 (grants no. 2010CB530301 and 2010CB530303) and the US National Institutes of Health Comprehensive International Program of Research on AIDS–affiliated Project (grant no. 3 U19 AI051915-05S1). D.L. is partially supported by the Natural Science Foundation of China (grant no. 30800640). G.F.G. is leading principal investigator of the Innovative Research Group of the National Natural Science Foundation of China (grant no. 81021003).

**Xudong Hu,[1] Di Liu,[1] Mingyang Wang, Le Yang, Ming Wang, Qingyu Zhu, Laixing Li, and George F. Gao**

Author affiliations: China Agricultural University College of Veterinary Medicine, Beijing, People's Republic of China (X. Hu, Ming Wang, G.F. Gao); Chinese Academy of Sciences (CAS) Key Laboratory of Pathogenic Microbiology and Immunology Institute of Microbiology, Beijing (X. Hu, D. Liu, Mingyang Wang, G.F. Gao); Graduate University CAS, Beijing (Mingyang Wang, G.F. Gao); Northwestern Institute of Plateau Biology CAS, Xining, China (L. Yang, L. Li); Tibet Plateau Institute of Biology, Lhasa, Tibet Autonomous Region, China (L. Yang); State Key Laboratory of Pathogens and Biosecurity Academy of Military Medical Sciences, Beijing (Q. Zhu); and Beijing Institutes of Life Sciences CAS, Beijing (G.F. Gao)

DOI: 10.3201/eid1703.100948

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[1]These authors contributed equally to this article.

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Address for correspondence: George F. Gao, CAS Key Laboratory of Pathogenic Microbiology and Immunology, Institute of Microbiology, Chinese Academy of Sciences, Beijing 100101, People's Republic of China; email: gaof@im.ac.cn



## Diagnosis and Treatment of Tuberculosis in the Private Sector, Vietnam

**To the Editor:** In many countries, the private sector (practitioners not employed by government and nongovernment institutions, e.g., hospitals, pharmacies) is a major source of care, even for poor persons, and the area where services for the public are widely available (*1*,*2*). However, little information is available from high-incidence countries about the role of the private sector in tuberculosis (TB) detection and treatment (*3*). In Vietnam, ≤40% of all TB cases in Ho Chi Minh City (the largest city in Vietnam and with the highest rate of economic growth in the country) were estimated to be treated in the private sector (*4*), and half of all patients with a diagnosis of TB in the public sector (National Tuberculosis Program [NTP]) in Ho Chi Minh City initially sought help in the private sector (*5*). However, this estimate does not reflect private care in the entire country.

In 2006–2007, a countrywide TB prevalence survey was conducted in Vietnam (*6*) in which data were obtained for previous TB treatment. This survey provided an opportunity to calculate a nationally representative estimate of the proportion of TB cases treated in the private sector and to investigate demographic characteristics of persons choosing treatment in this sector.

The study was reviewed and approved by the Research Board of the Vietnam National Lung Hospital. Details of survey methods have been reported (*6*). All eligible persons were screened to identify suspected cases of TB by using a short, structured, screening questionnaire and chest radiograph. Persons with suspected TB were those who reported persistent productive cough, who had radiographic abnormalities suggestive of TB, or who received TB treatment either currently or in the 2 years preceding the survey. Persons had an in-depth interview that included questions on where they were treated for TB. Assessment of socioeconomic status was based on 9 household characteristics (*7*).

Missing data were imputed by using multiple imputation methods, assuming that these data were missing at random to adjust for nonparticipation and missing data on facility of TB treatment (*8*). We used the ice and mi commands in Stata version 11 software (StataCorp LP, College Station, TX, USA), which included age, area, zone, and socioeconomic status.

Of the 103,924 eligible persons in selected districts, 94,179 (91%) were screened, 7,498 were identified as having suspected TB, and 407 reported having been recently treated for TB: 316 (77.6%) in public health facilities (PHFs) reporting cases to the NTP, 8 (2.0%) in PHFs not reporting cases to the NTP, and 29 (7.1%) in private health care facilities not reporting to the NTP. Fifty-four (13.3%) did not provide information about where they were treated. Multiple imputation led to adjusted proportions of 88.9%, 2.9%, and 8.2%, respectively. Sensitivity analyses, which assigned 54 persons with missing data for location of TB treatment to PHFs or private clinics, resulted in a range of 7.1%–20.3% for private sector treatment.

Characteristics of participants by type of facility where they received TB treatment are shown in the Table. Women, younger persons, and residents of southern Vietnam were more likely to seek treatment in the private sector. Urban populations and those with the highest socioeconomic status were most likely to seek private care, but these differences were not significant (Table).